

# Amphiphilic Block Copolymers Based on Methoxy Poly(ethylene glycol) and Either Crystalline or Amorphous Poly(caprolactone-*b*-lactide): Synthesis, Solid-State and Aqueous Solution Characterizations

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**ABSTRACT:** Amphiphilic block copolymers consisting of methoxy poly(ethylene glycol) (mPEG) and either crystalline poly(caprolactone-*b*-L-lactide) [P(CL-LLA)] or amorphous poly(caprolactone-*b*-D,L-lactide) [P(CL-DLLA)], respectively, were prepared under mild conditions using dicyclohexylcarbodiimide as coupling agent. Micelles were formed from the resultant copolymers with predetermined hydrophobic and hydrophilic blocks in water by dialysis method. The composition and structure of the copolymers were characterized by <sup>1</sup>H-NMR, <sup>13</sup>C-NMR, and GPC measurements. Differential scanning calorimetry analysis revealed that the crystallization behavior of P(CL-LLA) block is affected significantly by the molecular weight of the conjugated mPEG block, and the glass transition temperature (*T*<sub>g</sub>) of P(CL-DLLA) block is depressed by the existence of mPEG moiety in the block copolymer. The crystallization

behavior was also characterized by the technique of polarized light microscopy. The hydrodynamic diameter and size distribution of micelles were determined by particle size measurements. Transmission electron microscopy images showed that P(CL-LLA)-*b*-mPEG block copolymer self-aggregated into thread-like shape in water, whereas P(CL-DLLA)-*b*-mPEG adopted a classical spherical shape. It is suggested that both the high enthalpy of crystallization and hydrophobicity of P(CL-LLA) core-forming blocks would be responsible for the thread-like morphology. A possible mechanism for the thread-like assembly morphology was also discussed. © 2007 Wiley Periodicals, Inc. *J Appl Polym Sci* 105: 915–927, 2007

**Key words:** amphiphilic; block copolymers; methoxy poly(ethylene glycol); micelles; poly(caprolactone-*b*-lactide)

## INTRODUCTION

During the past decades, extensive efforts have been devoted to the development of amphiphilic block copolymers and their applications in biomedical fields.<sup>1–10</sup> It is well known that when amphiphilic block copolymers are dissolved in a solvent selective for one of the blocks, micelles are usually formed with a rather dense core of the insoluble blocks, surrounded by the diffuse outer corona formed from the soluble blocks. Block copolymers possessing hydrophobic polyester segments and hydrophilic polyether segments constitute a main class of micellar carriers for drug delivery. In the family of aliphatic polyesters, poly(L-lactide) (PLLA), poly(D,L-lactide) (PDLLA), poly(ε-caprolactone) (PCL), and their copolymers have received considerable attention as biocompatible and biodegradable polymers.<sup>11</sup> Poly(ethylene glycol) (PEG) is mostly selected as the polyether segments based on its excellent water-solubility and nontoxicity.<sup>12</sup>

Zhu and his coworkers synthesized poly(D,L-lactide)-*b*-PEG and evaluated its use as drug carrier.<sup>13,14</sup> Synthesis of poly(D,L-lactide)-*b*-methoxy poly(ethylene glycol) (mPEG) diblock copolymers was also described by Churchill and Hutchinson.<sup>15</sup> The diblock copolymer was synthesized from D,L-lactide and mPEG by ring opening polymerization in the presence of stannous octoate. Ring opening polymerization with similar reaction conditions has also been employed in the synthesis of polyester-mPEG diblock copolymers, where the polyester unit was poly(caprolactone) (PCL)<sup>16,17</sup> or poly(glycolic acid) (PGA)<sup>18</sup> and their copolymers<sup>19</sup>. In all cases, the polyester segments were introduced via ring opening polymerization initiated by the terminal hydroxyl group on the mPEG chain. Recently, Bae et al.<sup>20</sup> reported the synthesis of multiblock copolymers composed of short blocks of poly(ethylene oxide) (PEO) and poly(ε-caprolactone) or poly(L-lactide) by esterification of dicarboxylated PEO and dihydroxy PCL for PEO/PCL multiblocks, and dihydroxy PEO and dicarboxylated PLLA for PEO/PLLA multiblocks.

In this article, we report the synthesis and characterization of a series of block copolymers composed of mPEG as the hydrophilic segments and the diblock copolymers of poly(caprolactone-*b*-L-lactide)

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[P(CL-LLA)] or poly(caprolactone-*b*-D,L-lactide) [P(CL-DLLA)] as the hydrophobic segments, emphasizing especially on the predetermined molecular structure and diverse physical properties of the hydrophobic segments. Namely, the diblock copolymers of P(CL-LLA) and P(CL-DLLA) were selected on the aim of modulating the crystallinity and the glass transition temperature of the hydrophobic segments. In aqueous solution, the resultant amphiphilic block copolymers can self-assemble into well-defined micelles. Therefore, it is expected that the self-assembly behavior and accordingly the micellar characteristics as carriers for drug delivery will be influenced by the core-forming blocks with different crystalline nature.

## EXPERIMENTAL PART

### Materials

Methoxy poly(ethylene glycol) (mPEG) with molecular weights of 1100, 2000, and 5000 were purchased from Fluka. The materials were dried in vacuum at 60°C for 24 h before use. Dicyclohexylcarbodiimide (DCC) (Acros) was distilled under diminished pressure (b. p. 130°C/3 mmHg) prior to use. 4-(*N,N*-dimethylamino) pyridine (DMAP) and succinic anhydride were recrystallized from ethyl acetate and acetic anhydride, respectively. Triethylamine (TEA) was dried with 4A molecular sieve before use. 1,4-dioxane and methylene chloride were dried over sodium and anhydrous calcium chloride, respectively, and then distilled. All other chemicals were used as received.

P(CL-LLA) ( $M_{n(\text{GPC})} = 6614$  g/mol, PDI = 1.6) and P(CL-DLLA) ( $M_{n(\text{GPC})} = 8121$  g/mol, PDI = 1.4) diblock copolymers were kindly provided by Prof. R. Jerome and Dr. P. Lecomte in the University of Liege (Belgium). The copolymers were prepared by using  $\text{Bu}_2\text{Sn}(\text{OMe})_2$  as initiator, and caprolactone was polymerized first followed by the polymerization of lactide. One of the chain-ends of the diblock copolymers was capped with methyl ester group and the other end with hydroxyl group. LLA/CL and DLLA/CL molar ratios were calculated from the integration ratio of the bands due to PLA blocks at 5.1 ppm and to PCL blocks at 2.3 ppm to be 10.3/1 and 11.2/1 by  $^1\text{H-NMR}$  measurements, respectively.

Dialysis Tubing (Benzoylated cellulose) (MWCO: 1200) was purchased from Sigma-Aldrich Co. (USA), and Membra-Cel<sup>TM</sup> regenerated cellulose membrane (MWCO : 3500 and 7000) was from Serva (Germany).

### Preparation of mPEG bearing carboxylic end-group

Carboxylic acid-terminal mPEG was prepared according to Zalipsky et al.<sup>21</sup> and Bae et al.<sup>20</sup>. Briefly, succinic anhydride (3.6 mmol) and DMAP (3 mmol) were dissolved in 1,4-dioxane (10 mL) in a sealed

flask at room temperature. The flask was evacuated three times and filled with nitrogen. Then, dried mPEG (3 mmol) in 1,4-dioxane (30 mL) and TEA (3 mmol) in a small volume of 1,4-dioxane was added successively. The reaction was continued for 24 h at room temperature with stirring and nitrogen atmosphere protection. At the completion of reaction, dioxane was removed in a rotary vacuum evaporator. The residue was dissolved in  $\text{CCl}_4$  (30 mL) and the solid was filtered off. The filtrate was precipitated in diethyl ether. After filtration the product was dried in vacuum.

### Polyesterification reaction

For the synthesis of P(CL-LA)-*b*-mPEG block copolymers, carboxylic acid-terminated mPEG (0.4 mmol), P(CL-LA) (0.2 mmol), and DMAP (0.04 mmol) were dissolved in methylene chloride (30 mL) in a sealed flask. Under the same anhydrous condition, DCC (0.4 mmol) in methylene chloride (10 mL) was added to the previous flask. The reaction was continued for 24 h at room temperature. Precipitated dicyclohexylurea was removed by filtration. The filtrate was precipitated in diethyl ether, and the product was collected by filtration, then dried in vacuum at 40°C.

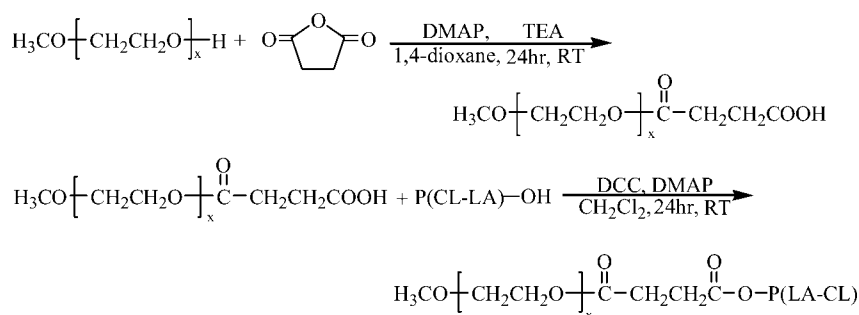
### Purification process

Unreacted mPEG moiety was removed from the block copolymer product by dialysis method. Typically, the product mixture was first dissolved in dimethylformamide (DMF) at 40°C. The solution was then cooled to room temperature and filtered if necessary. Then the solution was transferred to a dialysis tube (MWCO: 1200, 3500, and 7000, respectively, depending on the molecular weight of mPEG) to remove the unreacted mPEG. The products were lyophilized after dialysis.

In the following of the text, P(CL-LLA) and P(CL-DLLA) diblock copolymers are abbreviated as C-L and C-DL, respectively. P(CL-LLA)-*b*-mPEG and P(CL-DLLA)-*b*-mPEG block copolymers are abbreviated as (C-L)- $E_x$  and (C-DL)- $E_x$ , and the subscript numbers are the indicative molecular weight of the corresponding mPEG block in hundred g/mol.

### Preparation of micellar solutions

Because the micelles cannot be prepared by direct dissolution of the synthesized block copolymers in water, the modified dialysis method of Zhang and Eisenberg<sup>8</sup> were used. Typically, the block copolymers were first dissolved in *N,N*-dimethylformamide (DMF), a common solvent for both P(CL-LA) and mPEG blocks. Subsequently, deionized water was added to the polymer/DMF solutions (10 mL) at a



**Scheme 1** Synthetic route for P(CL-LA)-*b*-mPEG block copolymer.

rate of 1 drop every 10 s with vigorous stirring. The addition of water was continued until the water content reached 20–30 wt % depending on the composition of the block copolymers. The resulting solutions were transferred to a dialysis tube (MWCO: 1200, 3500, and 7000, respectively, depending on the molecular weight of mPEG) and dialyzed against doubly distilled water to remove the organic solvent at 4°C. After dialysis, many systems turned slightly cloudy, except for (C-DL)-E<sub>20</sub> and (C-DL)-E<sub>50</sub>, which remained transparent.

### Characterization

Nuclear magnetic resonance (<sup>1</sup>H-NMR, <sup>13</sup>C-NMR)

<sup>1</sup>H-NMR and <sup>13</sup>C-NMR spectra were recorded on an AVANCE DMX 500 MHz spectrometer at room temperature. Samples were dissolved in deuterated chloroform CDCl<sub>3</sub> containing tetramethylsilane.

### Gel permeation chromatography

Molecular weights were obtained by GPC with a Polymer Laboratories PL-GPC 220 Chromatograph, equipped with three PLgel columns at 40°C. Tetrahydrofuran was used as mobile phase at a flowing rate of 1 mL/min, and calibration was based on polystyrene standard.

### Differential scanning calorimetry

Differential scanning calorimetry (DSC) experiments were performed using a PERKIN ELMER Pyris 1 DSC. mPEG with molecular weights of 1100, 2000, 5000, P(CL-DLLA) and P(CL-DLLA)-*b*-mPEG block copolymers were analyzed by the following process: heated from 40 to 100°C at 20°C/min, kept at 100°C for 3 min, cooled to –70°C at 10°C/min (second run), and finally reheated from –70°C to 100°C at 10°C/min (third run).

P(CL-LLA) and P(CL-LLA)-*b*-mPEG block copolymers were analyzed using the following program: heated from 40 to 170°C at 20°C/min, kept at 170°C for 3 min, cooled to –70°C at 10°C/min (second run),

and reheated from –70°C to 170°C at 10°C/min (third run).

Then the crystallization temperature (*T<sub>c</sub>*) and melting temperature (*T<sub>m</sub>*) were taken as the peak values of the exotherms and endotherms from the second and third run, respectively.

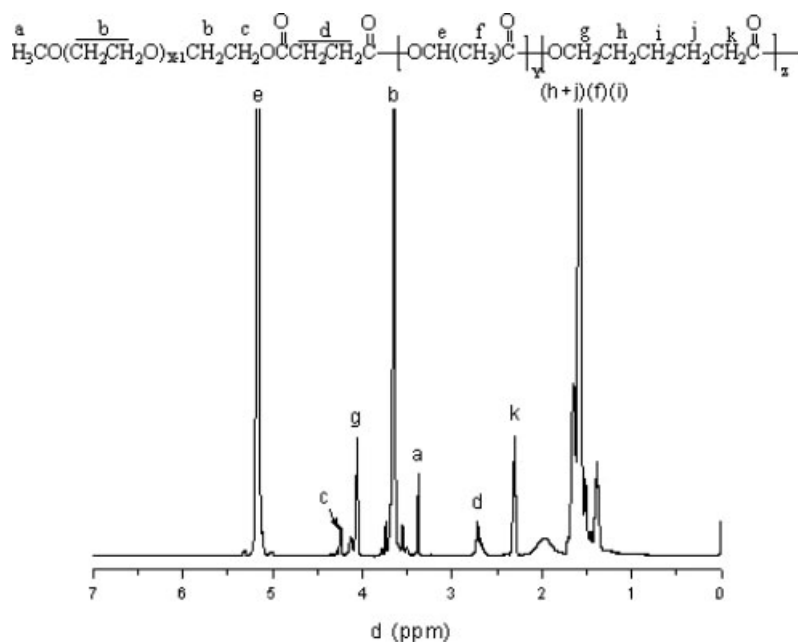
P(CL-LLA) and P(CL-LLA)-*b*-mPEG block copolymers identified as quenched were first heated up to 170°C and kept for 5 min, and then cooled down to –60°C. At a heating rate of 10°C/min, the samples were heated from –60°C to 170°C. The glass transition temperature (*T<sub>g</sub>*) was taken from the midpoint of heat capacity changes.

### Polarized light microscopy

The crystallization behavior of P(CL-LA), mPEG, and P(CL-LA)-*b*-mPEG block copolymers were analyzed by Olympus polarized light microscopy (PLM) equipped with a programmable Linkam css450 hot stage. The samples of P(CL-LLA) and P(CL-LLA)-*b*-mPEG were completely melted at 180°C and held for about 5 min, then cooled at a cooling rate of 5°C/min to 150°C, and then cooled at a rate of 0.5°C/min until the crystalline morphology appeared, and held at this temperature for 30 min. The samples of mPEG (MW: 1100, 2000, and 5000), P(CL-DLLA), and P(CL-DLLA)-*b*-mPEG block copolymers were melted at 100°C and held for 5 min, then cooled at a cooling rate of 5°C/min to 50°C, and then cooled at a rate of 0.5°C/min until the crystalline morphology appeared, and held at this temperature for 30 min.

### Particle size measurements

The hydrodynamic diameter and size distribution of micelles were determined by dynamic light scattering (DLS) using a Brookhaven 90 Plus particle size analyzer. Each analysis lasted for three 3 min and performed at 25°C with an angle detection of 90°. All micelle solutions had a final polymer concentration of about 0.1 mg/mL. Prior to the light scattering measurements the sample solutions were filtered through 0.45 μm filters.



**Figure 1**  $^1\text{H}$  NMR spectrum of P(CL-LLA)-*b*-mPEG block copolymer (C-L)-E<sub>11</sub> in  $\text{CDCl}_3$ .

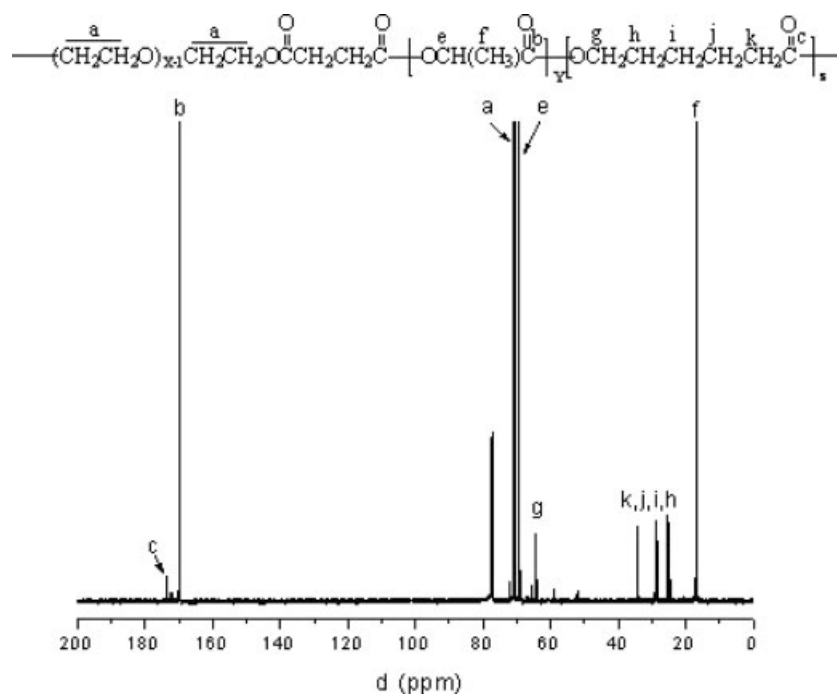
#### Transmission electron microscopy

Transmission electron microscopy (TEM) was performed on a JEOL JEM-1230 electron microscope operating at an acceleration voltage of 60 KV. Samples were deposited from micelle solutions onto copper grids coated with carbon. Water was evaporated from the grids at 10°C under normal atmospheric pressure. After that, the grids were negative stained by 2 wt % phosphotungstic acid.

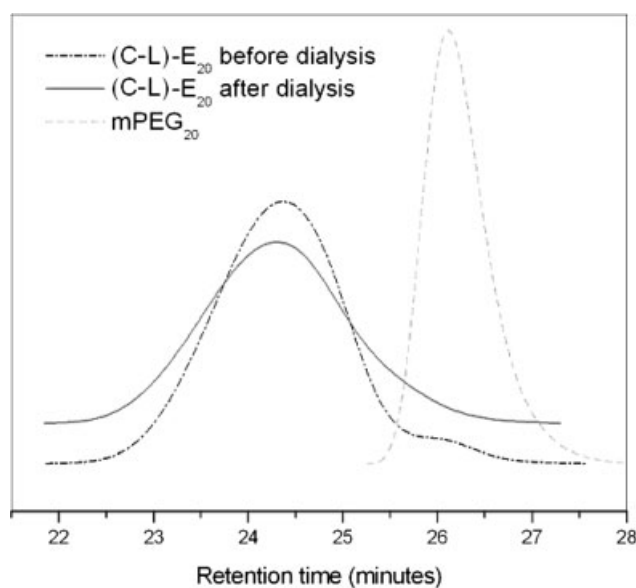
## RESULTS AND DISCUSSION

#### Synthesis and characterization

P(CL-LLA)-*b*-mPEG and P(CL-DLLA)-*b*-mPEG amphiphilic block copolymers were prepared at room temperature by reacting carboxylic acid-terminated mPEG and hydroxyl-terminated P(CL-LLA) or P(CL-DLLA) in the presence of DCC as coupling agent. The procedure is demonstrated in Scheme 1. Along this



**Figure 2**  $^{13}\text{C}$  NMR spectrum of P(CL-LLA)-*b*-mPEG block copolymer (C-L)-E<sub>11</sub> in  $\text{CDCl}_3$ .



**Figure 3** GPC chromatographs of P(CL-LLA)-*b*-mPEG block copolymer (C-L)-E<sub>20</sub> (before and after dialysis) and mPEG homopolymer.

way, a series of block copolymers with predetermined mPEG and P(CL-LLA), P(CL-DLLA) blocks were synthesized. The method gives a convenient way to architect flexibly the predetermined amphiphilic block copolymers.

The resultant block copolymers are characterized by NMR spectrum. Figure 1 shows the <sup>1</sup>H-NMR spectrum of (C-L)-E<sub>11</sub> block copolymer in CDCl<sub>3</sub>. The characteristic resonance peaks of PLLA, PCL, and mPEG components are obviously exhibited. The resonance peaks at 1.5 ppm (CH<sub>3</sub>) and 5.17 ppm (CH) are assigned to PLLA block, and the peaks at 1.3, 1.6, 2.3, and 4.0 ppm belong to the methylene protons (CH<sub>2</sub>) in PCL blocks (see details in Fig. 1). Signals at 3.6 ppm (CH<sub>2</sub>) and 3.37 ppm (OCH<sub>3</sub>) are the character of mPEG blocks.

Additional information of the chemical structure of P(CL-LLA)-*b*-mPEG block copolymer is obtained from <sup>13</sup>C-NMR spectrum (Fig. 2). In comparison with

the spectra of mPEG and P(CL-LLA), it shows clearly the signals at 16.9 ppm (CH<sub>3</sub>), 69.2 ppm (CH), 169.7 ppm (CO) corresponding to PLLA segments, and 70.7 ppm (CH<sub>2</sub>) to mPEG segments. In addition, resonance peaks at 24.6, 25.7, 28.3, 34.3, 64.3 ppm (CH<sub>2</sub>) and 173.8 ppm (CO) are the evidence of PCL segments within the polyester blocks in the copolymers.

The molecular weights of P(CL-LA), mPEG precursors and the resultant block copolymers were characterized by GPC analysis. It can be seen in Figure 3 that the initially bimodal chromatogram of the resultant copolymer becomes a unimodal profile after dialysis, indicating that the unreacted mPEG can be efficaciously separated from the system by dialysis method. Combined with the NMR data earlier, the results demonstrate that mPEG and P(CL-LA) are covalently bonded, forming the structure-predetermined block copolymers by the mild reaction process described previously. The molecular characteristics of the resultant block copolymers are shown in Table I.

#### Solid-state thermal behavior of P(CL-LLA)-*b*-mPEG (crystalline-*b*-crystalline) block copolymers

It is known that phase behavior in the solid state will show dramatic differences when conjugating two diverse blocks into one polymer chain, which may also affect significantly the assembly properties of the copolymers in solution. In an attempt to gain insight into the self-assembly behavior of amphiphilic block copolymers both in bulk and in solution, DSC study was carried out to get a perspective on the solid-state thermal properties of the resultant copolymers under a nonisothermal condition.

DSC thermograms of P(CL-LLA) and P(CL-LLA)-*b*-mPEG copolymers in the second heating run after a rapid cooling from the melt are shown in Figure 4. The quenched P(CL-LLA) copolymer shows a clear glass transition at 30°C, followed by a distinct cold crystallization exotherm located at 67.6°C. A melting endotherm of PLLA presents subsequently. No transition evidence relating to the PCL block exhibits dur-

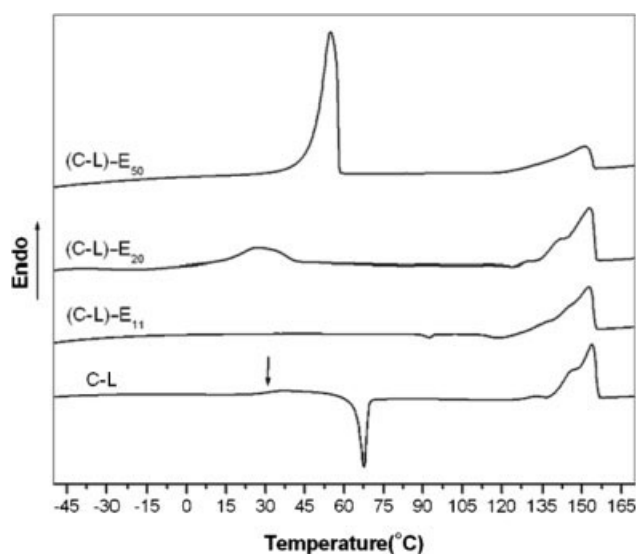
**TABLE I**  
Molecular Characteristics of the P(CL-LA)-*b*-mPEG Block Copolymers

Samples	Prepolymers		P(CL-LA)- <i>b</i> -mPEG				Hydrophobic block <sup>c</sup> (wt %)
	mPEG ( $M_n$ ) <sup>a</sup>	P(CL-LA) ( $M_n$ ) <sup>a</sup>	$M_n$ <sup>a</sup>	$M_w$ <sup>a</sup>	$M_w/M_n$ <sup>a</sup>	$M_n$ <sup>b</sup>	
(C-L)-E <sub>11</sub>	1146	6614	8882	12501	1.41	7949	89.6
(C-L)-E <sub>20</sub>	1997	6614	8939	13409	1.50	8287	82.2
(C-L)-E <sub>50</sub>	5408	6614	10301	14422	1.40	9771	54.5
(C-DL)-E <sub>11</sub>	1146	8121	9307	11679	1.25	5393	84.5
(C-DL)-E <sub>20</sub>	1997	8121	9497	11966	1.26	6421	76.6
(C-DL)-E <sub>50</sub>	5408	8121	10366	13268	1.28	8306	44.1

<sup>a</sup> Measured by GPC analysis.

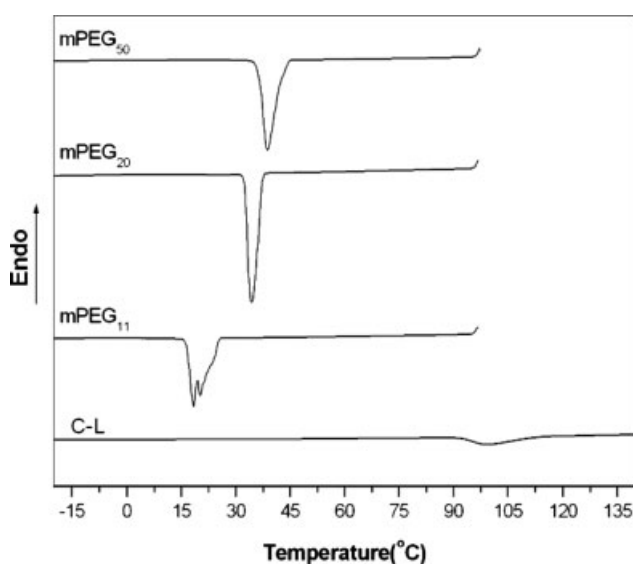
<sup>b</sup> Measured by <sup>1</sup>H-NMR measurements.

<sup>c</sup> Determined on the basis of combination of <sup>1</sup>H-NMR and GPC results.



**Figure 4** DSC heating curves of the quenched P(CL-LLA) and P(CL-LLA)-*b*-mPEG block copolymers.

ing quenching or the subsequent heating process. This may be attributed to the low content of PCL and the covalent bonding to PLLA in P(CL-LLA). For P(CL-LLA)-*b*-mPEG block copolymers with different mPEG block length, the  $T_g$ s of the copolymers cannot be observed and the cold crystallization of PLLA completely diminishes, only for (C-L)-E<sub>11</sub> that a very small cold crystallization peak presents. Combining mPEG block to P(CL-LLA) increases significantly the crystallization rate of PLLA moiety. It is clear that PLLA segments crystallize rapidly during the quenching procedure, resulting in less amorphous content and the disappearance of the  $T_g$ . Furthermore, for the copolymers of (C-L)-E<sub>20</sub> and (C-L)-E<sub>50</sub>, the

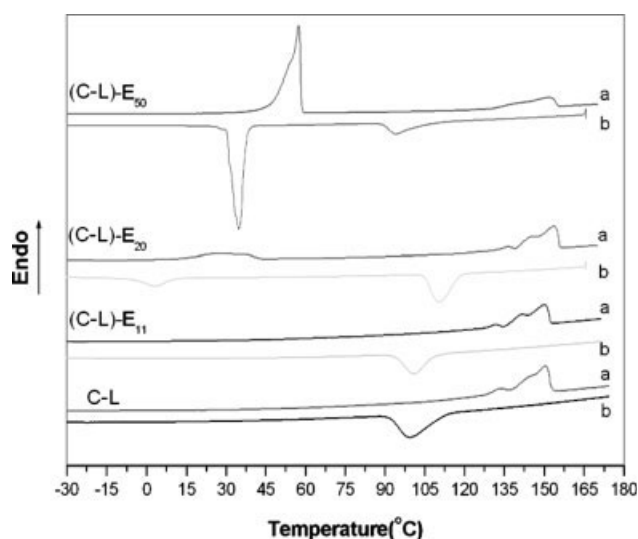


**Figure 5** DSC curves of the prepolymers cooling from the melt (second run).

glass transition temperatures of PLLA segments might be overlapped by the melting peaks of mPEG segments, which is in consistent with the literature data on PLLA/PEG blends.<sup>22</sup>

Figure 5 shows the crystallization curves of P(CL-LLA) and mPEG prepolymers cooled from the melt. P(CL-LLA) prepolymer exhibits one crystallization peak at  $\sim 99.5^\circ\text{C}$  assigned to PLLA. Since the crystallization temperature ( $T_c$ ) of mPEG homopolymers increases from 18 to  $38^\circ\text{C}$  with the increase of molecular weight from 1100 to 5000. It suggests that microphase separation may take place in P(CL-LLA)-*b*-mPEG copolymers, forming PLLA and mPEG microdomains when the copolymers are cooled from the melt. However, taking into account the covalent link between the two crystalline blocks, the restriction effect may suppress the mobility and accordingly the rearrangement of the individual block. On the other hand, an opposite effect to the restriction is the plasticizing effect of PEG on PLLA chains, which can enhance significantly the mobility of the chains.<sup>22-27</sup> Therefore, competitions between these opposite driving forces may dictate the phase behavior of these crystalline-*b*-crystalline copolymers when they are subjected to the cooling-reheating cycle at a constant rate.

The cooling-reheating curves of P(CL-LLA)-*b*-mPEG block copolymers with different compositions are illustrated in Figure 6. When (C-L)-E<sub>20</sub> and (C-L)-E<sub>50</sub> block copolymers are cooled from the melt, P(CL-LLA) and mPEG blocks crystallize independently at very different temperatures. In contrast, only one crystallization peak at  $101^\circ\text{C}$  corresponding to P(CL-LLA) segments is recorded for the copolymer (C-L)-E<sub>11</sub>. No crystallization of mPEG<sub>11</sub> segments can be



**Figure 6** DSC reheating curves (third run) (a) and cooling curves (second run) (b) of P(CL-LLA)-*b*-mPEG block copolymers and P(CL-LLA).

**TABLE II**  
Calorimetric Data of the P(CL-LLA)-*b*-mPEG Block Copolymers and Precursors

Sample	$T_c$ (°C) <sup>a</sup>		$\Delta H_c$ (J/g) <sup>b</sup>		$T_m$ (°C) <sup>c</sup>		$\Delta H_m$ (J/g) <sup>b</sup>		$T_{app}$ (°C) <sup>d</sup>
	mPEG	C-L	mPEG	C-L	mPEG	C-L	mPEG	C-L	
mPEG <sub>11</sub>	18.4		-162.5		40.5		168.1		31.8 ± 1.1
mPEG <sub>20</sub>	34.3		-171.5		52.8		176.3		43.4 ± 1.9
mPEG <sub>50</sub>	38.8		-180.6		59.2		185.4		45.8 ± 0.2
C-L		99.5		-43.4		150.5		41.6	127.3 ± 0.4
(C-L)-E <sub>11</sub>		101.0		-43.8		150.0		45.8	121.3 ± 0.6
(C-L)-E <sub>20</sub>	3.0	110.5	-61.7	-44.0	27.5	153.5	76.4	47.1	114.5 ± 1.3
(C-L)-E <sub>50</sub>	34.8	94.2	-158.8	-32.3	57.3	151.4	161.9	33.8	107.6 ± 0.7

<sup>a</sup> Determined from DSC (second cooling run from the melt at 10°C/min)

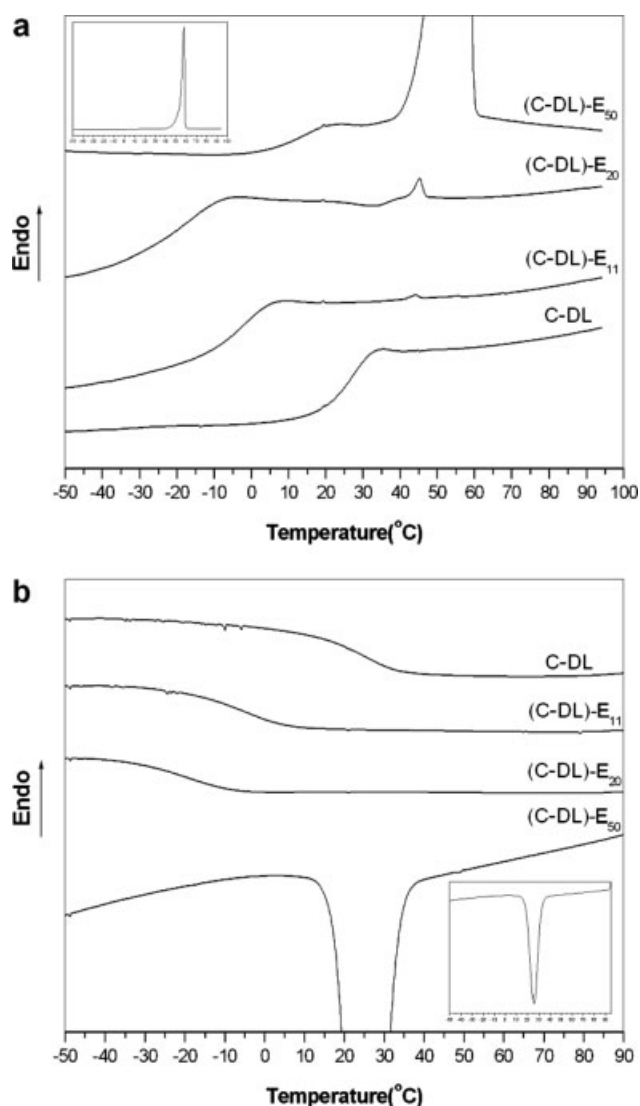
<sup>b</sup> Normalized values with respect to the wt % composition,  $\Delta H_{m(c)} = \Delta H_i/w_i$ , where  $\Delta H_i$  is the area of the endothermic or exothermic peak of mPEG or P(CL-LLA) block recorded from DSC thermograph, and  $w_i$  is the weight fraction of the corresponding block.

<sup>c</sup> Determined from DSC (third reheating run at 10°C/min).

<sup>d</sup> Temperature when the crystal structure appears determined by the technique of PLM equipped with a hot stage.

detected even after an annealing treatment. The result implies that the arrangement of mPEG<sub>11</sub> segments in the block copolymer becomes quiet difficult when it is covalently bonded to P(CL-LLA) block. A similar phenomenon is found in the reheating curves of the copolymers. Two melting peaks exhibit in the DSC thermograms of (C-L)-E<sub>20</sub> and (C-L)-E<sub>50</sub>. The lower melting peaks at 27 and 57°C are assigned, respectively, to mPEG<sub>20</sub> and mPEG<sub>50</sub> blocks. The subsequent multimodal melting peak at about 150°C is attributed to P(CL-LLA) block as a result of lamellar reorganization during PLLA crystallization.<sup>25,26</sup> No melting sign for mPEG<sub>11</sub> but only one endothermal peak of P(CL-LLA) block demonstrates in the thermogram of (C-L)-E<sub>11</sub>.

Thermal transitions and enthalpies of the block copolymers, mPEG and P(CL-LLA) prepolymers are listed in Table II. It can be found that the melting and crystallization enthalpies of P(CL-LLA) blocks in the copolymers of (C-L)-E<sub>11</sub> and (C-L)-E<sub>20</sub> increase slightly as compared with that of the pure P(CL-LLA) prepolymer, indicating that the crystallinity of P(CL-LLA) blocks is enhanced rather than depressed by the presence of mPEG with relatively low molecular weights. Whereas, the enthalpy of P(CL-LLA) blocks in (C-L)-E<sub>50</sub> is apparently lowered. Despite the changes in the melting enthalpies, the melting temperatures of the corresponding P(CL-LLA) blocks are little affected. In fact, as the copolymers are cooled from the melt, P(CL-LLA) blocks crystallize first, and at this stage, mPEG segments remain the molten state. It is reasonable to suppose that liquidlike mPEG chains with low molecular weights (i.e., mPEG<sub>11</sub> and mPEG<sub>20</sub>) penetrate into the amorphous region of P(CL-LLA) blocks, and behave similarly as plasticizer, resulting in the enhanced chain flexibility of P(CL-LLA). Consequently, reorganization of the polymer chains within the crystalline PLLA phase is favored to improve the perfection of PLLA crystals,



**Figure 7** DSC reheating curves (third run) (a) and cooling curves (second run) (b) of P(CL-DLLA)-*b*-mPEG block copolymers and P(CL-DLLA). The inset is an enlarged view of (C-DL)-E<sub>50</sub> sample.

**TABLE III**  
**Calorimetric Data of the P(CL-DLLA)-*b*-mPEG Block Copolymers and Precursor**

Sample	$T_g$ (°C) <sup>a</sup>	mPEG		$T_g$ (°C) <sup>c</sup>	mPEG		$T_{app}$ (°C) <sup>d</sup>
		$T_c$ (°C) <sup>a</sup>	$\Delta H_c$ (J/g) <sup>b</sup>		$T_m$ (°C) <sup>c</sup>	$\Delta H_m$ (J/g) <sup>b</sup>	
C-DL	20.1			26.0			–
(C-DL)-E <sub>11</sub>	–6.4			–3.7	44.2	0.57	–
(C-DL)-E <sub>20</sub>	–21.1			–18.6	45.2	2.31	29.6 ± 0.5
(C-DL)-E <sub>50</sub>		25.6	–238.1	12.3	57.8	244.4	37.0 ± 0.4

<sup>a</sup> Determined from DSC (second cooling run from the melt at 10°C/min).

<sup>b</sup> Normalized values with respect to the wt % composition,  $\Delta H_{m(c)} = \Delta H_i/w_i$ , where  $\Delta H_i$  is the area of the endothermic or exothermic peak of mPEG block recorded from DSC thermograph, and  $w_i$  is the weight fraction of the corresponding block.

<sup>c</sup> Determined from DSC (third reheating run at 10°C/min).

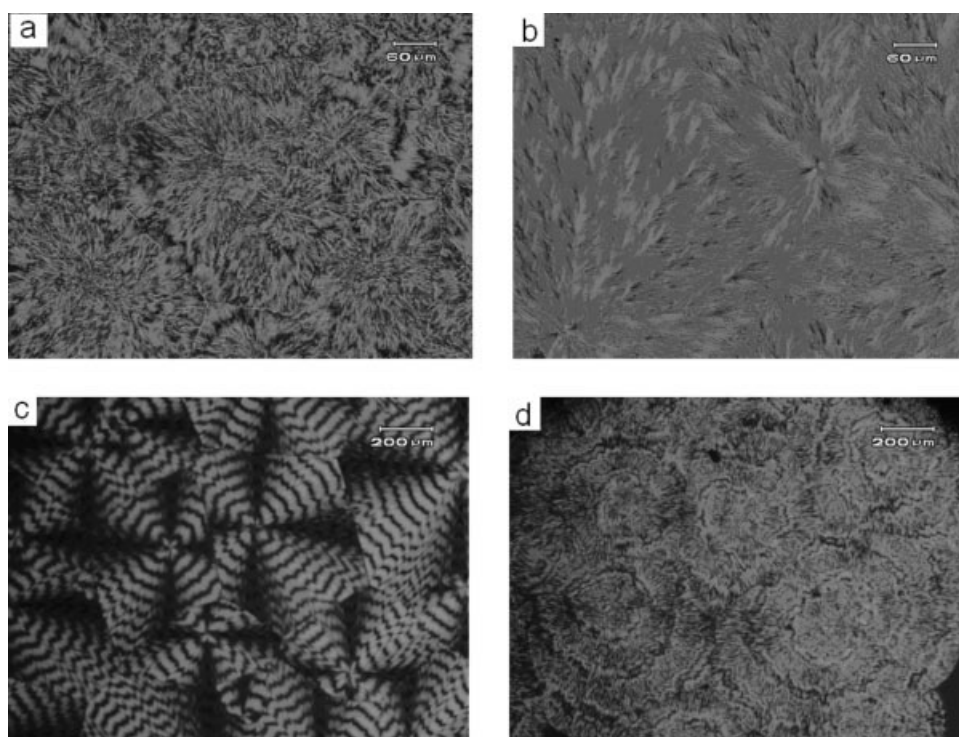
<sup>d</sup> Temperature when the crystal structure appears determined by the technique of PLM equipped with a hot stage.

as reflected by the slight increase in the  $\Delta H$  values of P(CL-LLA) block. Herein, the plasticizing effect of mPEG on P(CL-LLA) blocks plays a dominant role over the covalent bonding restriction between the two blocks. On the other hand, decreased melting and crystallization enthalpies of mPEG blocks in the copolymers of (C-L)-E<sub>20</sub> and (C-L)-E<sub>50</sub> are shown together with the decline of thermal transition temperatures, comparing with the corresponding mPEG homopolymers. The depression is even greater for mPEG in (C-L)-E<sub>20</sub> copolymer. This effect is considered to be an indication that some mPEG segments are trapped between PLLA crystals and the chain mobility is strongly subjected to the restriction of the earlier crystallized P(CL-LLA) block. Hence, the crystallization of mPEG<sub>11</sub> and mPEG<sub>20</sub> segments is sup-

pressed distinctly. When the molecular weight of mPEG and P(CL-LLA) blocks becomes equal in (C-L)-E<sub>50</sub> (see Table I), the mutual interference and restriction between the two blocks result in depressed crystallization for each block.

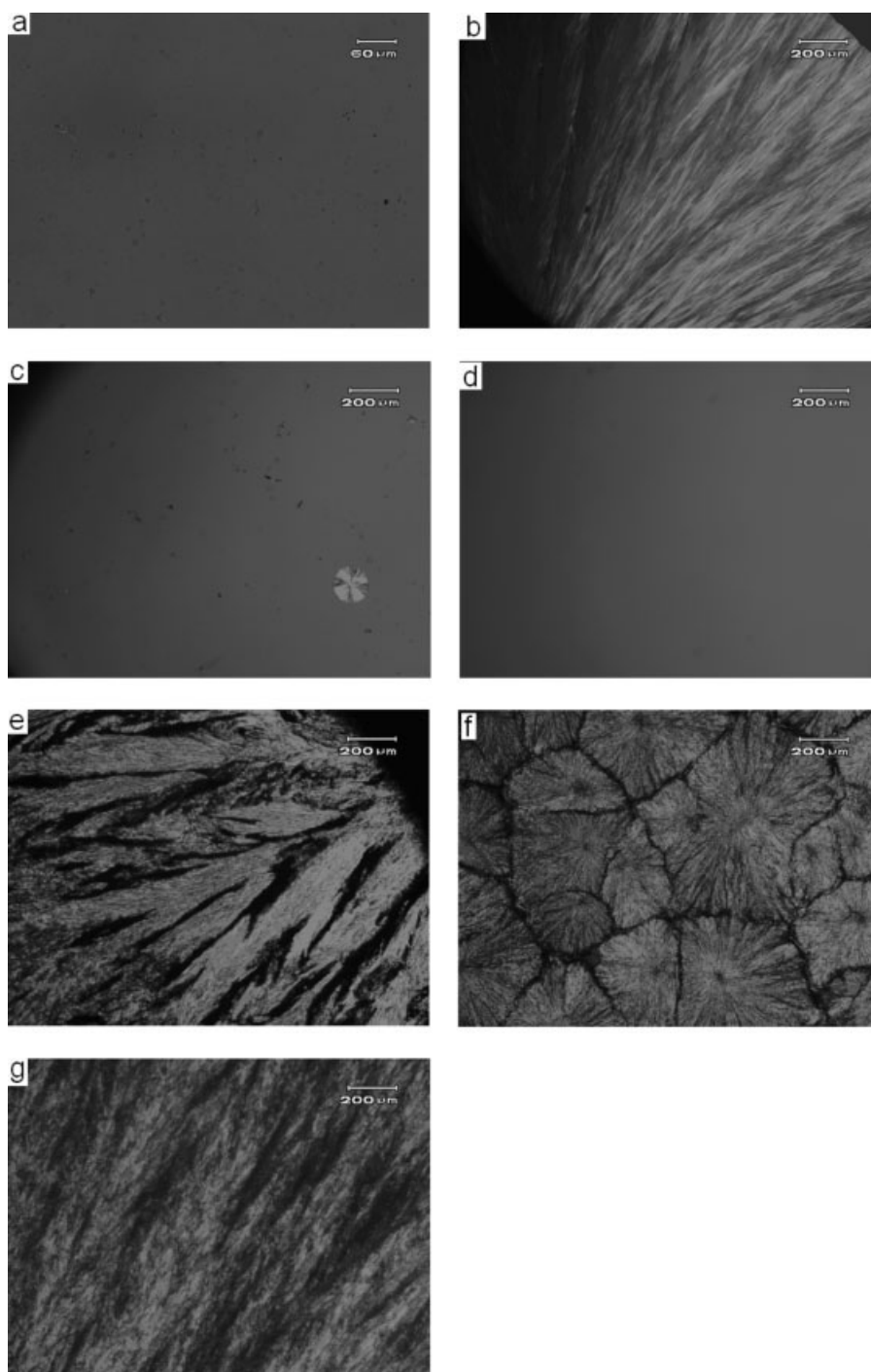
#### Solid-state thermal behavior of P(CL-DLLA)-*b*-mPEG (amorphous-*b*-crystalline) block copolymers

Since P(CL-DLLA) block copolymer exhibits an amorphous property, the thermograms of P(CL-DLLA)-*b*-mPEG block copolymers are expected to reveal different relaxation information contrasting to P(CL-LLA)-*b*-mPEG block copolymers. In the heating thermograms of P(CL-DLLA) precursor and P(CL-DLLA)-*b*-mPEG copolymers [Fig. 7(a)], one can find that the



**Figure 8** Optical micrographs of (a) P(CL-LLA); (b) (C-L)-E<sub>50</sub>; (c) (C-L)-E<sub>20</sub>; (d) (C-L)-E<sub>11</sub> block copolymers after crystallizing for 30 min at a certain temperature.

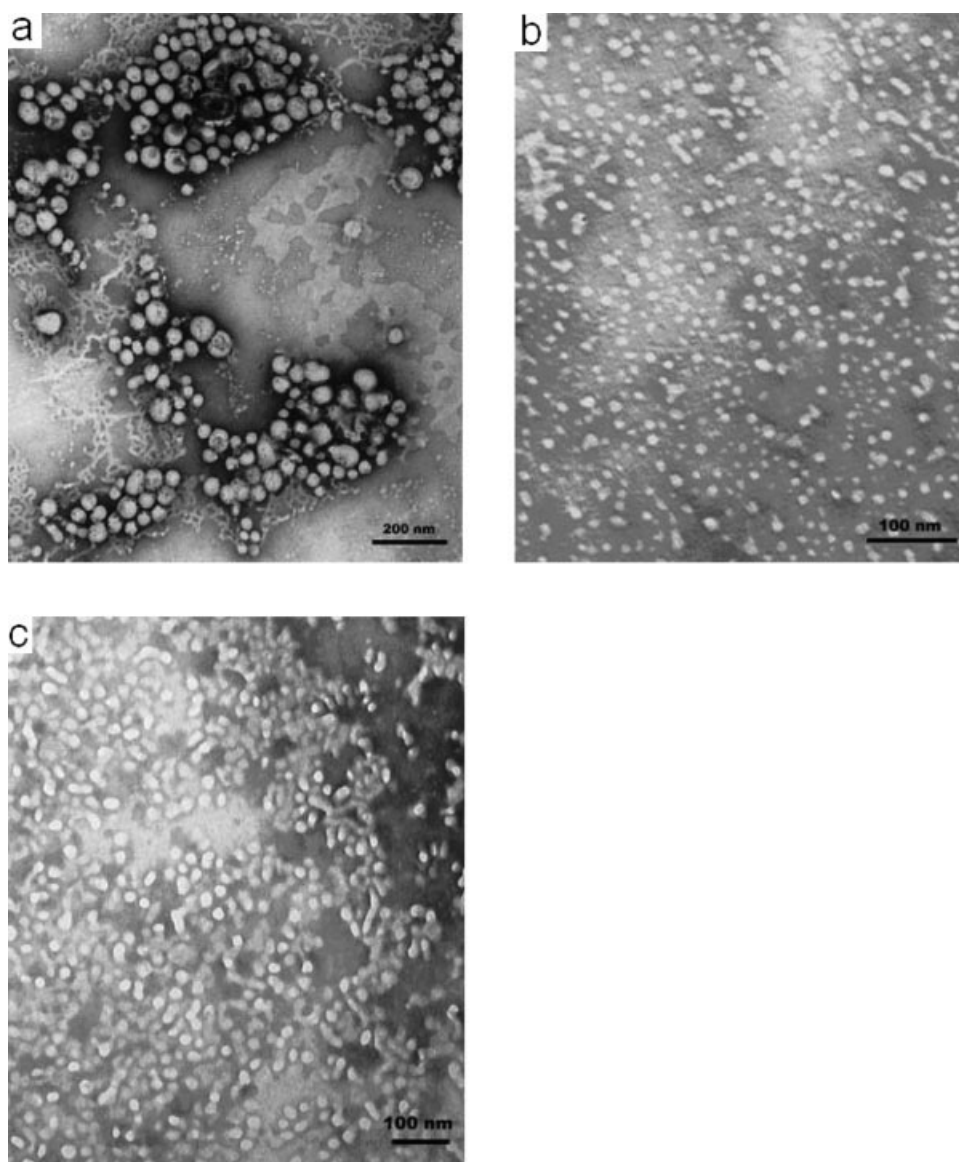




**Figure 9** Optical micrographs of (a) P(CL-DLLA); (b) (C-DL)-E<sub>50</sub>; (c) (C-DL)-E<sub>20</sub>; (d) (C-DL)-E<sub>11</sub>; (e) mPEG<sub>50</sub>; (f) mPEG<sub>20</sub>; (g) mPEG<sub>11</sub> block copolymers after crystallizing for 30 min at a certain temperature.

glass transition temperature ( $T_g$ ) of the amorphous P(CL-DLLA) precursor is located at about 26.0°C. Increasing the length of mPEG block in P(CL-DLLA)-*b*-mPEG block copolymers from 1100 to 2000 induces a visible decrease in the  $T_g$ s of P(CL-DLLA) block from -3.7°C to -18.6°C. The result is indicative of the miscibility between mPEG and P(CL-DLLA) blocks, which is further evidenced by the almost completely suppressed crystallization of mPEG<sub>11</sub> and mPEG<sub>20</sub>

blocks in the corresponding copolymers [Fig. 7(b)]. However, for the (C-DL)-E<sub>50</sub> block copolymer the glass transition temperature of P(CL-DLLA) segments shifts oppositely to higher temperature. It is conceivable that the result is related to the extraordinarily high degree of crystallinity of mPEG<sub>50</sub> within (C-DL)-E<sub>50</sub> copolymer. The melting enthalpy of the mPEG<sub>50</sub> segments in the copolymer is 244.4 J/g (Table III), which is dramatically higher than that of mPEG<sub>50</sub>



**Figure 10** TEM images of spherical micelles from P(CL-DLLA)-*b*-mPEG block copolymers: (a) (C-DL)-E<sub>11</sub>; (b) (C-DL)-E<sub>20</sub>; (c) (C-DL)-E<sub>50</sub>.

block in P(CL-LLA)-*b*-mPEG copolymer and that of mPEG<sub>50</sub> homopolymer, presumably because the amorphous P(CL-DLLA) block acts as a nucleus during the crystallization process of mPEG<sub>50</sub> block.

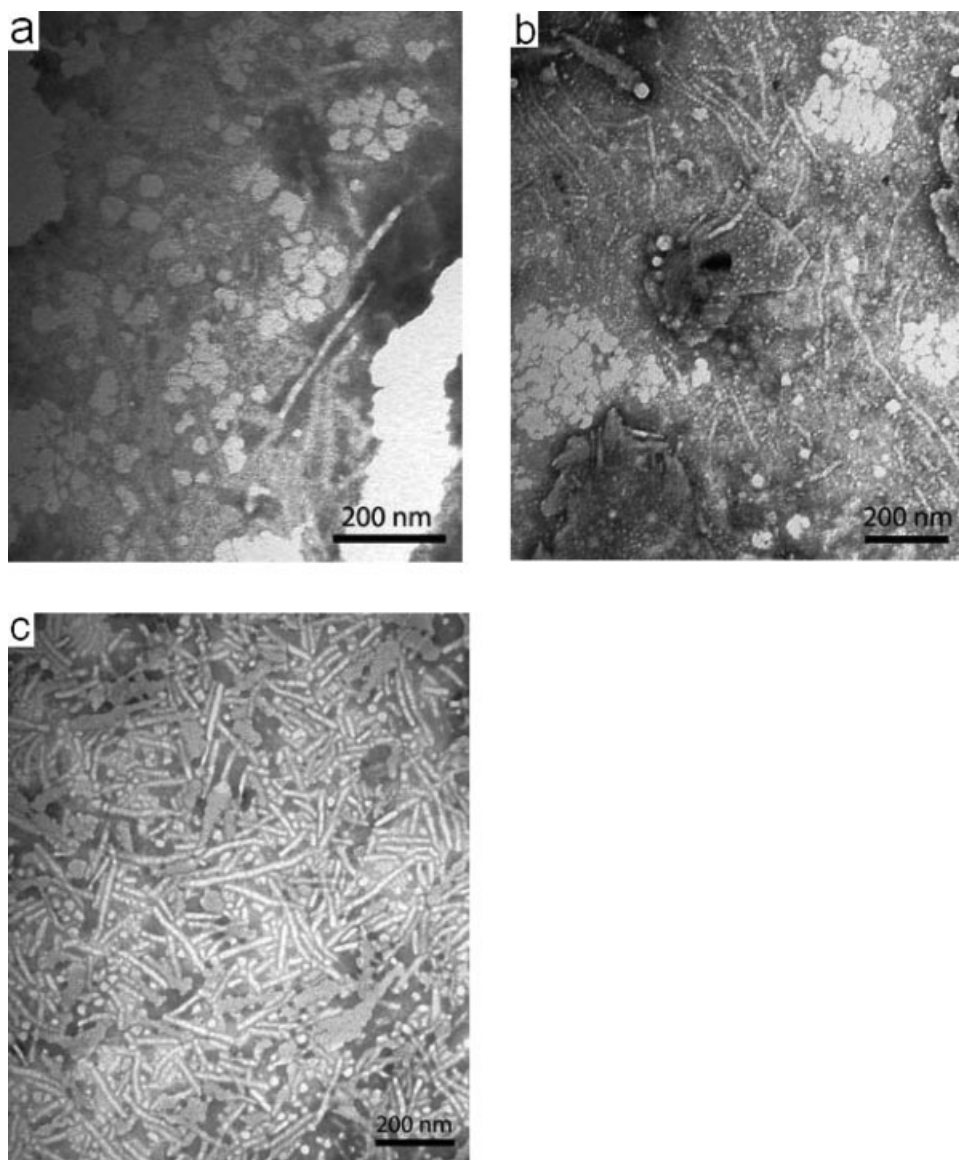
### Polarized light microscopy

Figure 8 exhibits the optical micrographs of the P(CL-LLA) and P(CL-LLA)-*b*-mPEG block copolymers. All the samples show the typical black cross under PLM. After 30 min at a certain temperature, the spherulites all have overlaid the slide. But with the increase of mPEG block length, the observed temperatures when PLLA spherulites appear were depressed (Table II). The results evidently indicate the depression effect of mPEG block on the mobility of PLLA chains. Similarly, in Figure 9, the crystallization of mPEG chains

was also suppressed obviously by the P(CL-DLLA) block (also shown in Table III). Especially in the case of (C-DL)-E<sub>11</sub>, even after the copolymer was held at 29°C for 30 min, the crystallization was not observed.

### Hydrodynamic diameters in aqueous solution

When amphiphilic block copolymers are dissolved in a solvent selective for one of the blocks, micelles can form as a result of the association of the insoluble blocks. It has been known that the more hydrophobic of micellar core is, the more copolymer chains are aggregated into a micelle to minimize the interfacial energy. Therefore, the size of the constructed micelles is larger. In agreement with this principle, one can find from the hydrodynamic diameters of the micelles formed by (C-DL)-E<sub>11</sub> is larger than those of (C-DL)-



**Figure 11** TEM images of thread-like micelles from P(CL-LLA)-*b*-mPEG block copolymers: (a) (C-L)-E<sub>11</sub>; (b) (C-L)-E<sub>20</sub>; (c) (C-L)-E<sub>50</sub>.

E<sub>20</sub> and (C-DL)-E<sub>50</sub> (Fig. 12). With the increase of hydrophilic mPEG block length and thus the elevated stability, the aggregation number of the copolymers into a micelle decreases, resulting in the decrease in micellar size.

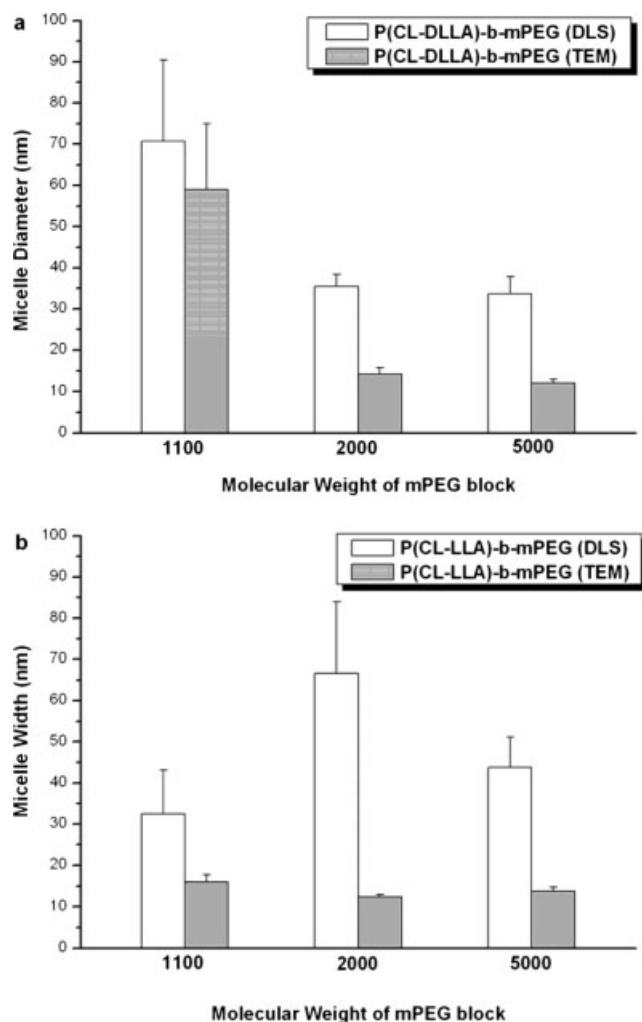
However, for the micelles constructed from P(CL-LLA)-*b*-mPEG copolymers with three different mPEG block lengths, the micellar sizes do not seem to follow the principle above.

### Micelle size and morphology

In this study, TEM technique is employed to image the size and shape of the formed micelles. The morphology of the micelles made from P(CL-DLLA)-*b*-mPEG copolymers are illustrated in Figure 10a. It can be seen that the micelles adopt a spherical shape and

keep the same morphology over the range of investigated mPEG length. The size values estimated by TEM are relatively smaller than those obtained from the DLS measurements (Fig. 12). This is believed to be due to the presence of water in the DLS experiments, which inducing the swelling of the hydrated mPEG corona. However, it is important to note that the size-changing trend in TEM images is in good agreement with the results in DLS measurement. Generally, the size of micelles decreases as the hydrophilic mPEG block length increases. So it is inferred from this result that the molecular weight of hydrophilic block plays a dominant role on the size of the micelles derived from P(CL-DLLA)-*b*-mPEG.

In contrast, the micelles constructed by P(CL-LLA)-*b*-mPEG copolymers exhibit a thread-like shape. A typical TEM image is shown in Figure 11. The width



**Figure 12** (a) Micelle diameter of P(CL-DLLA)-*b*-mPEG as well as (b) micelle width of P(CL-LLA)-*b*-mPEG from TEM and DLS measurements as a function of the molecular weight of hydrophilic mPEG block.

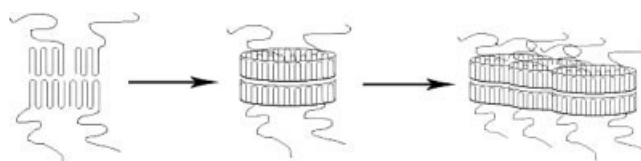
of the thread-like micelles in all cases is found to be  $\sim 10$ – $20$  nm, with lengths varying from 50 nm to several hundred nanometers. The increase of mPEG block length does not affect the width of micelles so visibly as in the case of P(CL-DLLA)-*b*-mPEG (Fig. 12b). It is worth to note that the size of the thread-like micelles measured by TEM cannot be directly compared with that obtained from DLS, since the basic equations to calculate the hydrodynamic diameter ( $d$ ) in DLS measurement are originated from the correlation function cumulant analysis, which is based on the assumption that the particles are noninteracting spheres and not anisotropic objects.

Inferring from the diverse physical nature of hydrophobic P(CL-LA) blocks in solid state confirmed by DSC earlier, we propose that both the high enthalpy of crystallization and hydrophobicity of P(CL-LLA) core-forming blocks would be responsible for the thread-like micellar morphology.

### Proposed mechanism for the thread-like morphology

The scaling model of Vilgis and Halperin (VH) provides a theoretical foundation in understanding the formation of thread-like micelles in this study.<sup>28</sup> VH consider a crystalline-coil A-B diblock copolymer that aggregates in a highly B-selective solvent, forming micelles with a chain-folded crystalline core and swollen corona. Because of the incompatibility of A-B blocks in highly selective solvent, both core and corona chains are considered to be grafted to the core-corona interface. High surface tension in the core-solvent surface favors more copolymers to be aggregated into one micelle to decrease the surface area per copolymer. But as the micelles become larger, the densely grafted corona chains are more stretched because of an increase of the repulsion interaction. Consequently, it leads to a free energy penalty and thus restricting the aggregates' growth. The micellar equilibrium structure is determined by a balance between these two terms: the surface free energy associated with the core-solvent interface and the free energy penalty due to the stretching of the densely grafted corona blocks.

For the present P(CL-LLA)-*b*-mPEG copolymers, as described earlier, the micelles are prepared by first dissolving the block copolymers in DMF, and subsequently slowly adding water to induce micelle formation, i.e., the aggregation of the P(CL-LLA) core blocks. At the initial stage of water addition, the quality of the solvent for P(CL-LLA) core blocks decreases slowly. At this stage, the system is both thermodynamically and kinetically controlled. When the content of water exceeds some critical value and finally the aggregates are isolated into water by dialysis, both of the lowered chain mobility and high enthalpy of crystallization of P(CL-LLA) core blocks make the system become kinetically inaccessible. Though in crystalline cores the configuration of the crystallizable core blocks is adjustable, there is no free energy change related with the configurational adjustments because the chain packing is ordered. Figure 13 shows a schematic illustration of the mechanism for micellization with thread-like morphology. It involves a transition originated from starlike crystalline-coil micelle, which exhibits a double stack of folded crystallized core surrounded by extended corona blocks.



**Figure 13** Schematic illustration of the micellization with thread-like morphology in solution.

The final formation of thread-like micelles is consistent with a strong competition between the energy of the crystalline core and chain stretching of the densely grafted mPEG corona. Because of the high surface tension between the hydrophobic core and water, more densely grafted mPEG chains is favored to a flat grafted layer to decrease the average area per corona chain, that is to say, a transition from a sphere to a lamellar or thread-like morphology would be preferred. But the entropy loss from the stretching of corona chains in the case of lamellar therefore will be excessively considerable. A thread-like structure provides the best balance between these effects.

### CONCLUSIONS

Amphiphilic P(CL-LLA)-*b*-mPEG and P(CL-DLLA)-*b*-mPEG block copolymers with different compositions were synthesized under mild conditions by reacting carboxylic acid-terminated mPEG with hydroxyl-terminated P(CL-LLA) and P(CL-DLLA) in the presence of DCC as coupling agent. The <sup>1</sup>H-NMR, <sup>13</sup>C-NMR, and GPC analyses confirmed the formation of block copolymers. Modulating the predetermined block length of mPEG, P(CL-LLA), and P(CL-DLLA) allowed the flexible tailoring of the molecular architecture of the amphiphilic copolymers.

DSC investigation showed that the molecular weight of mPEG block in the copolymers can significantly influence the thermal properties of the hydrophobic P(CL-LLA) and P(CL-DLLA) blocks in the solid state. The PLM observations also validated these results. The crystallinity of P(CL-LLA) blocks in (C-L)-E<sub>11</sub> and (C-L)-E<sub>20</sub> increases compared with the P(CL-LLA) precursor. However, the crystallinity of P(CL-LLA) blocks in (C-L)-E<sub>50</sub> is suppressed apparently. The *T*<sub>g</sub>s of P(CL-DLLA) blocks in copolymer (C-DL)-E<sub>11</sub> and (C-DL)-E<sub>20</sub> decrease with increasing mPEG block length, whereas the *T*<sub>g</sub> of P(CL-DLLA) blocks in (C-DL)-E<sub>50</sub> approaches to that of P(CL-DLLA) precursor. The crystallization of mPEG blocks in these copolymers is restricted because of the covalent bond between P(CL-LA) and mPEG blocks, except for (C-DL)-E<sub>50</sub>, wherein the crystallinity of mPEG<sub>50</sub> block is extraordinarily enhanced.

Because of the amphiphilic nature of such block copolymers, they can spontaneously self-assemble into well-defined micelles in aqueous solution. TEM images showed that micelles made from P(CL-DLLA)-*b*-mPEG copolymers adopted a spherical shape and kept the same morphology over the range

of investigated mPEG length. In contrast, the micelles arising from P(CL-LLA)-*b*-mPEG copolymers exhibited a thread-like shape. Our results suggest that the thread-like micelles may be owed to the crystallization of P(CL-LLA) core-forming blocks in the course of micellization. We propose a possible mechanism for the thread-like morphology of P(CL-LLA)-*b*-mPEG micelles: a strong competition between the energy of the crystalline core and chain stretching of the densely grafted mPEG corona would be responsible for the formation of thread-like micelles.

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